1998

Status Report

Stored-Product Insects Research of of Military Importance

Biological Research Unit
U. S. Grain Marketing Research Laboratory
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1998 Status Report

"Stored-Product Entomology Research"

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U. S. GRAIN MARKETING RESEARCH LABORATORY

GRAIN MARKETING AND PRODUCTION RESEARCH CENTER

LABORATORY MISSION

The mission of the U. S. Grain Marketing Research Laboratory is to develop new knowledge, information, and technologies needed to solve problems associated with the harvest, storage, marketing, and overall quality and end-use properties of cereal grains. The Laboratory program focuses on post-harvest aspects of grain marketing and includes a major commitment to incorporation of optimum end-use quality characteristics into new grain varieties in cooperation with plant breeders. The Laboratory provides multidisciplinary research on these issues by three research units:

Biological Research Unit

Engineering Research Unit

Grain Quality and Structure Research Unit

MISSION OF THE BIOLOGICAL RESEARCH UNIT

The mission of the Biological Research Unit is to develop new, ecologically-based methods to manage insect pests in stored grain, processed commodities and storage and processing facilities. A multi-disciplinary team conducts fundamental and applied research on new biological and ecological approaches to pest management that will reduce the use of traditional pesticides on grain and grain products. Successful completion of the mission will provide a sustainable supply of high-quality cereal products.

We conduct research on biological control agents, insect-resistant packaging, novel physiological control techniques, host plant resistance, insecticide deployment and resistance management strategies, insect biochemistry and genetics, insect population monitoring, population dynamics and behavior, and computer-based integrated pest management systems.

[illegible] RESEARCH LABORATORY

GRAIN MARKETING AND PRODUCTION RESEARCH CENTER

LABORATORY MISSION

The mission of the U.S. Grain Marketing Research Laboratory [illegible]

[illegible] Research Unit

Engineering Research Unit

Grain Quality and Structure Research Unit

MISSION OF THE [illegible] RESEARCH UNIT

The mission of the [illegible] Research Unit is to [illegible]

[illegible]

BIOLOGICAL RESEARCH UNIT
GRAIN MARKETING AND PRODUCTION RESEARCH CENTER

STAFFING/AREA OF RESEARCH EMPHASIS

Research Leader

Dr. James E. Throne Supervisory Research Entomologist	Ecology, Modeling, Seed Resistance to Insects

Scientists

Dr. Franklin H. Arthur Research Entomologist	Integrated Pest Management Technologies
Dr. James E. Baker, Research Entomologist	Parasitoid Biology and Toxicology, Digestive Physiology
Dr. Richard W. Beeman Research Entomologist	Genetics/Molecular Biology, Insecticide Resistance
Dr. Alan K. Dowdy Research Entomologist	Insect Ecology/Behavior in Commercial Facilities
Dr. Paul W. Flinn Research Biologist	Modeling/Expert Systems
Dr. David W. Hagstrum Research Entomologist	Insect Ecology, Modeling, Sampling, Acoustic Detection
Dr. Ralph W. Howard Research Chemist	Chemical Ecology, Biological Control
Dr. Karl J. Kramer Research Chemist	Insect Biochemistry, Physiology, Biopesticides
Dr. Jeffrey C. Lord Research Entomologist	Insect Pathology
Dr. Michael A. Mullen Research Entomologist	Insect Trapping, Insect Resistant Packaging
Dr. Brenda S. Oppert Research Biochemist	Insect Biochemistry, Physiology, Toxicology, *Bacillus thuringiensis* Research

RESEARCH HIGHLIGHTS AND TECHNOLOGY TRANSFER FOR FY98:

Stored Grain Advisor upgraded and made available to public. The model in the Stored Grain Advisor (SGA) expert system was programmed to predict the effects of using automatic aeration controllers starting at harvest. The KSU brochure on "Questions and answers about aeration controllers" was added to the SGA help. SGA was made available for free download from our web site at http://bru.usgmrl.ksu.edu/flinn. In the last 6 months over 150 copies have been downloaded. (Flinn)

Movement of grain beetle in temperature gradients analyzed and can now be predicted. We studied the movement of rusty grain beetle in a temperature gradient in grain, and found that this insect moves into warmer regions. These results will be incorporated into a spatial insect model that we have developed, and should make predictions by SGA more accurate. (Flinn)

Aeration of stored grain has major impact on efficiency of beneficial insects. Studies were conducted on the effects of early aeration vs. no aeration on efficacy of the parasitic wasp, *Choetospila elegans* to suppress lesser grain borer. This study should have a major impact on biological control in stored grain because it shows that the wasps are ten-times more effective in suppressing beetles at 25°C than at 32°C. (Flinn)

Biotic stresses attenuate insect responses to bacterial infection. Studies demonstrated that if insects had been previously exposed to bacterial challenges, then they have a lessened ability to fight off new infections. These findings suggest that control of insect pests with pathogens might successfully use multiple sublethal applications. (Howard)

Natural products that facilitate mating behavior on beneficial insects characterized. Studies demonstrated that the cuticular hydrocarbons of the parasitoid, *Cephalonomia tarsalis,* varied with gender, age, and nutritional- and mating-status, and that the females produce a sex pheromone. These chemicals are used by the insects to facilitate mating. Studies also demonstrated that the parasitic wasp, *Pteromalus cerealellae*, has a complex behavioral pattern that it uses for its courtship and mating behavior. These behaviors incorporate a sex pheromone and gender specific cuticular hydrocarbons which appear to be used for final gender recognition. This information will be used to develop improved biological control strategies that use this parasitoid. (Howard)

Automated spectroscopic method detects hidden insects. A near infrared spectrophotometer was integrated with the single kernel characterization system and used to rapidly and automatically detect wheat kernels infested with in-kernel feeding (hidden) insects. This method can be easily incorporated into the current grain grading system to provide information on insect infestations in raw grain. (Dowell/Throne/Baker)

Automated spectroscopic method detects parasitized weevil larvae. Parasitoid pupae inside grain kernels could be easily differentiated from weevil pupae, weevil larvae, and uninfested wheat kernels by use of a near infrared reflectance spectrophotometer. As part of an artificial rearing method for this parasitoid, this rapid method to differentiate parasitized from non-parasitized weevils in grain could have practical application for biological control companies releasing immature parasitoids into grain storage facilities. (Baker/Dowell/Throne)

Diatomaceous earth (DE) adversely impacts efficacy of beneficial insects in grain. DE, an inert dust insecticide, reduces the efficacy of a parasitic wasp attacking weevils in wheat. This finding has implications for integrating biological control with DE treatments for stored product insect pest control. (Perez/Baker/Arthur/Flinn)

Mechanism of insecticide resistance in beneficial wasp determined. Increased expression of a malathion-specific carboxylesterase is the major mechanism conferring resistance in a malathion-resistant parasitoid found in stored grain. Understanding development of resistance in parasitoids will facilitate integration of chemical and biological controls. (Baker/Fabrick/Zhu)

Genetic mutation responsible for insecticide resistance in beneficial wasp characterized. PCR Amplification of specific allelle was used to detect a single base pair substitution in cDNA from resistant and susceptible strains of a parasitic wasp. This base pair substitution results in an amino acid difference in the encoded protein from the two wasp strains, but we do not know how these structural differences may relate to functional differences in the protein. Genes conferring resistance can be incorporated into transformed beneficial insects. (Zhu/Dowdy/Baker)

Effects of seasonal changes in grain temperatures on trap catch populations evaluated. The influence of the seasonal changes in grain temperatures in two bins of stored wheat on each of two farms in Kansas on the prediction of insect density from probe trap catch was investigated, and equations were developed for converting probe trap catches for the four most common species to insect density per volume of grain. This research will make probe traps more useful as a monitoring tool for stored-grain pest management programs. (Hagstrum)

Incidence of resistance to natural insect toxins determined. The lack of a major digestive proteinase in the Indian meal moth was linked to resistance to the microbial toxins of *Bacillus thuringiensis.* Digestive gut proteinase patterns were characterized in geographically distinct populations of the moth to determine the extent of heterogeneity in proteinase activities. The prevalence of proteinase-mediated resistance to *Bacillus thuringiensis* toxins will influence the strategies used to manage *Bacillus thuringiensis*-resistance in field insects. (Oppert)

Resistance of stored corn to insect pests demonstrated. We showed that about half of 72 commercial corn hybrids tested were relatively resistant to maize weevils. The resistant hybrids would have a longer storage life than the susceptible hybrids. (Throne/Baker)

Gut proteinases of several stored grain beetle species evaluated. Stored product beetles varied in their response to beetle-active *Bacillus thuringiensis* toxins. Because serine proteinases are involved in the solubilization of beetle-active toxins, we examined whether the complement of gut proteinases was correlated to *Bacillus thuringiensis* toxicity among various stored product beetles. In beetles analyzed thus far, higher levels of serine proteinase activity were correlated with higher toxicity. This work will lead to more effective, environmentally safe *Bacillus thuringiensis*-based control products for stored product beetle pests. (Oppert/Kramer)

Mechanism of skeletal cross-linking reactions determined. Insects use highly reactive agents to cross-link proteins and stabilize their exoskeletons. Until now, the reactivity of these agents has not been understood. A unifying mechanism has been delineated, which predicts the regioselective outcomes and explains the product distribution of these reactions. Intramolecular base catalysis occurs when these agents cross-link proteins, which determines the nature of the cross-links formed in the cuticle. (Kramer)

Novel biopesticides for insect and fungal pests programmed into plants. Tobacco and corn plants have been genetically engineered to express novel insect control genes. Two specific examples of these genes code for chitinase and avidin. Chitinase is an enzyme that degrades structural components in the insect gut. Avidin is a protein that binds to the vitamin biotin and interferes with an insect's ability to produce energy from its food. Tobacco was used as a model system as a first step in developing these techniques in cereals because it is much easier to manipulate than wheat, corn, rice, etc. The expression of these genes alone or in combination with other defense proteins confers resistance to insect and fungal pests. Similar work is now being done in the more genetically complex cereal plants. (Kramer)

New method for distinguishing structures of modified proteins in natural systems developed. With cooperators at the University of Washington, we developed a facile method for distinguishing modified amino acids in biomaterials using electrospray ionization tandem mass spectrometry. In the past it has been very difficult to establish the precise structures of these products which are found in insects, marine animals, plants, and microorganisms, and also are associated with several neurological disease states. (Kramer)

New method for analysis of compounds diagnostic for the presence of insect pests in stored products developed. We developed a sensitive and precise method of analysis of compounds (quinones and hydroquinones) secreted by insects in stored grain and processed products. Theses compounds are used for defensive purposes by insects and their levels depend not only on the species, age and sex of the insects but also on the lability of the compounds extracted from the samples collected. (Kramer)

Automated spectroscopic method kills insects in grain. We killed all rice weevil larvae feeding inside of wheat kernels using near-infrared spectroscopy. (Dowell/Baker/Throne)

Automated spectroscopic method identifies adult stored-product insects. Using near-infrared spectroscopy we could classify adult stored-product insects as either primary or secondary pests with 99% accuracy, and also classify according to genus with 95% accuracy. (Dowell/Baker/Throne/Wang)

Fungal colonists of stored corn evaluated. We determined the fungal colonization of corn stored at seven constant temperatures, ranging from 10° to 40°C, and four constant moisture contents, ranging from 9 to 18%, over a 2-year period. Twenty fungal species were identified. *Eurotium chevalieri* colonized a high proportion of kernels stored at the higher moisture contents and displaced some preharvest fungi. (Throne)

Mechanisms of hybrid inviability elucidated. We found that killer genes known as "Medea" (M) factors are required for larval mortality associated with the hybrid inviability gene "H". Knowledge of the detailed mechanisms of larval kill in the M and H systems could be exploited in new, naturally-based insect control strategies. (Beeman)

New genes required for embryonic differentiation discovered. We developed a large-insect genomic library, and used it to clone a large cluster of about 10 genes needed to direct the process of early insect growth and development. Molecular genetic analysis of these genes will lead to a better understanding of mechanisms of insect development. (Beeman)

New method for cloning hard-to-get genes developed. We developed a map-based cloning strategy for cloning flour beetle genes for which no DNA sequence information is available. The strategy involves a combination of high-resolution recombinational mapping, and large-insert genomic library screening using bacterial artificial chromosomes. We are now applying this approach in attempts to clone maternal, insect-lethal "Medea" genes. (Beeman)

Insect-resistant packaging study completed. In a cooperative project with Tenneco Corp., studies were completed which led to the approval of methyl salicylate as a repellent treatment for packages to improve insect resistance. (Mullen)

Use of heat sterilization for insect pest management evaluated. We developed a method for evaluating the efficiency of heat sterilization for insect management in processing facilites. Improved heat distribution was found to reduce treatment costs and improve insect control. (Dowdy)

A combination treatment of heat sterilization and diatomaceous earth for insect control developed. We determined the efficacy of combining heat sterilization and diatomaceous earth for insect management in processing facilities. Lower temperatures were effective and treatment times were shortened by the combined treatment, thus reducing facility down time for insect management. (Dowdy)

Fall aeration of corn helps to control insect pests. We showed that fall aeration is beneficial in reducing insect population in stored corn, but a second aeration cycle in late spring may have little effect. (Arthur)

Insect pests controlled with pyrethroid insecticide. We determined exposure intervals of 24 hours or less may not be long enough to control rice weevils on wheat treated with cyfluthrin EC, but the lesser grain borer was killed if exposed for only 8 hours. We also showed that the residual efficacy of an encapsulated formulation of cyfluthrin was not affected when wheat was stored between 20-35°C and that flour beetles exposed on concrete treated with cyfluthrin recovered when provided with food. (Arthur)

Effectiveness of Different Insect Sampling Devices in Farm-Stored Wheat. Similar information on insect infestation level with which to make pest management decisions was obtained with different sampling devices, and knowledge of the insect infestation level in an adjacent bin or the same bin during a previous year could not replace sampling to determine insect density. (Hagstrum)

Area-Wide IPM for Suppression of Insect Pests in Stored Wheat. Insect infestation levels were measured and the cost and effectiveness of current pest management programs was evaluated in two segments of the wheat marketing system as a first step toward improving insect pest management and reducing their dependence upon pesticides. (Hagstrum)

Identified commercial oat cultivars that have resistance to storage insect pests. Found large differences in number of progeny produced and development time of insects developing on commercial oat cultivars. Some cultivars were almost totally resistant to storage insect pests. (Throne)

Time-mortality probit analysis program. Provided copies of and instructions for using a time-mortality probit analysis program that colleagues and I developed to over 100 scientists at government laboratories and universities throughout the world. (Throne)

Behavior of parasitoids of stored grain pests. Demonstrated that the parasitoid *Pteromalus cerealellae* has a complex behavioral pattern that it uses to locate and recognize its hosts. This information will be used to develop improved biological control strategies that use this parasitoid. (Howard)

Review article on eicosanoids. Co-authored a major review on the role of eicosanoids in invertebrate biology with Professor David Stanley, University of Nebraska. This review emphasizes that the insect eicosanoid immune system is a novel target for new pesticide development. (Howard)

Capillary electrophoresis technique developed for analysis of phenolic acids in corn. Ferulic acid, a resistance factor against insects in corn, was quantified in test cultivars. (Baker/Fabrick).

Low temperature treatments applied to parasitized weevils in wheat prevented host emergence while allowing parasitoids to emerge. This technique could be used by companies that produce biologicals to use augmentative release of immatures without the concomitant release of host insects. (Baker)

Page esterase patterns in malathion-resistant *Habrobracon hebetor* were significantly different from those of susceptible strains of this parasitoid. Although total esterase activity in the resistant strain was lower than that in the susceptible strain, PAGE results indicated that specific properties of esterases in the R strain may be implicated in the resistance. No differences in activity levels in R and S strains were found for five other detoxification systems . (Baker/Fabrick)

Multigene trypsin family found in the intestinal tract of the lesser grain borer. Biochemical assays with specific substrates and inhibitors were used to characterize the serine proteinases that are potential targets for transgenes incorporated into cereals. (Zhu/Baker)

HPLC/PAGE method developed to isolate and purify the malathion-specific carboxylesterase responsible for malathion resistance in *Anisopteromalus calandrae*. Amino acid sequence of the purified enzyme will be used to complete the genetic characterization of insecticide resistance in this parasitoid. (Baker/Fabrick)

Molecular basis of malathion resistance in the parasitoid *A. calandrae* studied. A point mutation as well as expression levels of a carboxylesterase, as revealed by Northern blots, may be responsible for the high malathion resistance in this wasp. (Zhu/Baker)

Collaborated with Exelixis Pharmaceutical Co. to plan research on gene mining in pest insects to develop new insect control proteins. New genetic pathways will be identified that are susceptible to pest control intervention, using sophisticated molecular genetic techniques now being developed in Tribolium. (Beeman)

Pigment gene in red flour beetle cloned. We cloned the entire white gene and cDNA, required for normal tissue pigmentation in insects. This gene will be used as a tag to identify foreign DNA insertions in genetically transformed insects. The technology will be useful for genetic modification and improvement of beneficial insects, for monitoring of pest or beneficial insect populations, and for developing new concepts and approaches for biology-based pest control. (Beeman)

Grant received for development of novel biopesticide for crop protection. A research grant of $190,000 was received from the USDA National Research Initiatives Competitive Grant Program to work on the improvement of chitinolytic enzymes as biopesticides in transgenic plants. (Kramer)

Knowledge of insect exoskeletal biochemistry may lead to more effective pest Management. The insect exoskeleton is a good target for novel pest management strategies because of the unique insect-specific chemistry that occurs during its formation and recycling. Development of exoskeleton-targeted insect control agents has been hampered by a lack of basic knowledge about insect skeletal structure and metabolism. ARS and university scientists have identified novel metabolites and reactions in insects, which help to assemble and disassemble the exoskeleton. Inhibition of these reactions by biopesticides may be an environmentally-safe method of insect pest control. (Kramer)

Synergy between diatomaceous earth and *Beauveria bassiana* discovered. True synergy was discovered between the entomogenous fungus *Beauveria bassiana* and diatomaceous earth in assays against the lesser grain borer. (Lord)

Review articles on pest insect biology and applications of biotechnology for pest control published. Unit scientists authored several book chapters and journal review articles on the use of biotechnology for insect pest management. These will assist the scientific community in their research projects and also pest control operators in controling pest insects. (BRU scientists)

Entomological research journals edited by several unit scientists. Several unit scientists are members of the editorial boards of entomological research journals (Insect Biochemistry and Molecular Biology and Journal of Economic Entomology) for which they evaluate and edit manuscripts submitted for publication by the scientific community at large. (BRU scientists)

Patent issued for biopesticide for crop protection. A notice of allowance was issued by the U.S. Patent and Trademark Office on April 28, 1998 for U.S. CPA Patent Application S/N 08/524,051 entitled RECOMBINANT CHITINASE AND USE THEREOF AS A BIOCIDE, Kramer et al., filed 09/06/95. Chitinase is an enzyme that degrades the polysaccharide chitin found in the guts and exoskeletons of insects and cell walls of fungi. Several crops are being genetically engineered to express the recombinant protein encoded by an insect chitinase gene for host plant resistance to insect and fungal pests. (Kramer)

RESEARCH PROGRESS AND PLANS

CRIS No. 5430-43000-014-00D

Title: Ecology, Modeling, and Integrated Management of Stored-Product Insects

Main Objectives:

Pest management decisions can be improved by developing better insect monitoring programs and models for predicting insect population growth. Research will provide the technology that grain managers need to detect insect infestations more accurately and earlier, predict insect population growth rates and effects of different control measures, and forecast when insect control will be needed. These management tools are needed for a transition to less chemically dependent integrated pest management programs. This project focuses on acquiring essential biological and ecological data, using these data to develop predictive models of insect population growth, and developing an expert system for stored grain management that uses the models. Studies will expand the range of species and conditions over which existing models predict insect population growth and the range of problems for which the expert system can make pest management recommendations. Expansions will include additional pest species, natural enemies, grain varieties, aeration, cold temperature survival, and insect movement. Improved sampling programs will be developed to collect ecological data and acquire information needed to make pest management decisions. Automatic insect monitoring using acoustical sensors will be investigated.

Investigators: **David W. Hagstrum, James E. Throne, Paul W. Flinn**

1. Specific Objective: Area-Wide IPM for Suppression of Insect Pests in Stored Wheat.

Progress FY98: During the 2nd year of a 5-year area-wide IPM demonstration project, data were collected on the type, efficacy and cost of insect pest management done in networks of 11 elevators in Kansas and 12 elevators in Oklahoma. Elevators were selected so that grain could be followed as it moved from farm to country elevator to terminal elevators and finally left the elevator network to be milled or exported. Two former elevator managers were hired to supervise the work. The majority of bins in Kansas were concrete and the majority of bins in Oklahoma were steel. Aeration equipment was inventoried in preparation for installing aeration controllers during the 3rd year of the study. Methods were developed for sampling grain for insects and technicians were trained. The steel bins were sampled using a vacuum probe. Grain being removed from concrete bins was sampled with a pelican sampler at a rate of 1 kg/100 bushels to detect insect infestations early. We will resample this grain to measure insect population growth rates and the efficacy of insect pest management. Grain temperature data were collected weekly from all of the bins with thermocouples.

Plan of Work for FY99: Aeration controllers will be installed to increase the effectiveness and reduce the cost of aeration. The cost and effectiveness of this modified pest management program will be compared with that of the previous year. Managers with be encouraged to use our insect monitoring data in making pest management decisions. A commercially practical insect monitoring program will be developed to provide managers with the information about insect infestation levels that they use in making pest management decisions.

2. Specific Objective: Effectiveness of Different Insect Sampling Devices in Farm-Stored Wheat

Progress FY98: Newly harvested wheat stored in each of two bins on each of two farms in Kansas was sampled during each of three years at two locations (in the center and midway between the center and bin wall) within each bin. Several different sampling devices were used to monitor insect subpopulations in the headspace above the grain, on the grain surface and within the grain mass. *Typhaea stercorea* (L.) were found mainly in the headspace and on the grain surface. *Rhyzopertha dominica* (F.) were found mainly within the grain mass. *Cryptolestes ferrugineus* (Stephens) and *Ahasverus advena* (Waltl) were more evenly distributed among the three locations. This distribution pattern of insects among locations was consistent throughout the storage period. Insect populations in stored wheat increased steadily until the grain began to cool rapidly in the fall. More insects were found in the center than midway between the center and the bin wall. Which of the two bins on a farm was most heavily infested varied with species and years. The contribution of farm, year, bin, and location within a bin to the overall variance for insect density differed with insect species and sampling device. The majority of variance (37 to 95%) was due to differences between samples taken at the same location within a bin. The variance component for years tended to be small. The largest variation in insect densities between farms was that for *C. ferrugineus*. The variations between bins in the densities of each of the other three species tended to be larger than those for *C. ferrugineus*. Variance between locations within a bin was large for *C. ferrugineus* with all sampling devices, and for the catch of all species in sticky traps. This study found that similar information on insect infestation level with which to make pest management decisions might be obtained with different sampling devices, and that knowing the insect infestation level in an adjacent bin or the same bin during a previous year might not eliminate the need for sampling to determine insect density.

Plan of Work FY99: The relationship between bin size or the number of other bins on a farm and the level of insect infestation in each bin will be studied.

3. Specific Objective: Determine the effects of augmentative release of parasitoid wasps in stored wheat on insect fragments in flour.

Progress FY98: Field studies were conducted to assess the effectiveness of the parasitoid wasp, *Theocolax elegans* (Westwood) for controlling *Rhyzopertha dominica* (lesser grain borer) and *Cryptolestes ferrugineus* (rusty grain beetle) in 6 bins, each containing 27 tons of wheat. We also investigated the effects of parasitoid augmentation on insect fragment counts in flour that was milled from the grain samples. Beetles were released at monthly intervals into

all 6 bins. Parasitoid wasps were released into 3 of the bins. Adult populations of *R. dominica* were reduced by about 90% compared to the control. *R. dominica* larvae develop inside wheat kernels and were probably the main source of insect fragments in the flour. In 1994, insect fragment counts were significantly lower in samples that came from bins in which parasitoids were released than in the control bins. Because of a relatively low infestation rate, fragment counts were not significantly different in 1993. Insect myosin (a better indicator of insect contamination than fragments) followed the same trends as insect fragments. In 1994, insect myosin was significantly lower in flour samples from the treatment compared to the control bins. The number of insect damaged kernels (IDK) was significantly lower in the treatment than in the control bins in both 1993 and 1994. In the control bins, the IDK level was above the FGIS threshold in both 1993 and 1994. This study showed that augmentative parasitoid releases do not increase the number of insect fragments in flour. On the contrary, the number of insect fragments was greatly reduced in flour milled from bins that were treated with parasitoid wasps compared to the control bins.

Plan of Work FY99: Develop and validate a model of *Theocolax elegans* and its *host Rhyzopertha dominica* (lesser grain borer).

4. Specific Objective: Determine the effects of temperature gradients in stored wheat on the movement and distribution of the lesser grain borer beetle.

Progress FY98: The lesser grain borer beetle, *Rhyzopertha dominica*, is one of the most common and damaging insect pests of stored wheat in the United States and Canada. Larvae develop and feed inside the kernel and cause considerable damage. Beetle population growth rate is primarily affected by grain temperature. In the autumn, the periphery of the grain mass cools more rapidly than the center. Beetle populations may be higher in the center if they are able to move from the cool periphery towards the warm center of a grain mass. Temperature gradients were established in a 56 cm diameter cylinder with 9 cm high sides filled with 19.9 kg of hard red winter wheat to determine if the rusty grain beetle would disperse to warmer areas. Results showed that the lesser grain borer did not move into the warmer areas of the grain mass after 24 hours. A previous study with the rusty grain beetle, *Cryptolestes ferrugineus* showed that it would move into warmer areas of the grain mass. The center of a large bin of unaerated grain will often remain warm during the winter months, which allows insect populations to continue to develop and reproduce. Because the lesser grain borer does not appear to move into warmer regions of a grain mass, its population growth rate may be less than other species that are able to move into warmer regions. However, cooling the grain with aeration to a temperature of 15°C or less would severely reduce population growth of all of the damaging insects that attack stored grain.

Plan of Work for FY99: Continue evaluation of other stored grain beetles to determine if they also move towards warmer areas of the grain mass.

5. Specific Objective: Determine the effects of temperature and life stage on phosphine-induced mortality for the lesser grain borer, rice weevil, and rusty grain beetle.

Progress FY98: A specific cooperative agreement was established with Dr. Thomas Phillips of Oklahoma State University. He has started to conduct experiments with the lesser grain borer.

Plan of Work for FY99: We will analyze the data from Dr. Phillips and develop models to predict the effects of phosphine-induced mortality for the lesser grain borer, rice weevil, and rusty grain beetle.

6. Specific objective: Determine use of near-infrared reflectance spectroscopy for detection of insect in grain kernels.

Progress FY98: Obtained funding to hire a postdoctoral research associate to continue work in 1999.

Plan of Work for FY99: Improve sensitivity of the NIRS system through studies to identify the components of the insect that are primarily responsible for detection of the insects and use this information to modify the NIR wavelengths that we use to scan grain kernels. We will extend this work to determine whether insect fragments in flour can be detected and quantified using NIRS, which would allow automation of a labor-intensive food inspection process.

7. Specific objective: Determine use of near-infrared reflectance spectroscopy for killing insects in grain kernels.

Progress FY98: We achieved complete mortality of rice weevils in wheat kernels after 90 sec exposure to NIR.

Plan of Work for FY99: No further studies planned.

8. Specific objective: Determine use of near-infrared reflectance spectroscopy for classification of insects and other organisms.

Progress FY98: We extended this work to determine the age of insects.

Plan of Work for FY99: We are completing studies to determine whether NIR can be used to determine the age of insects.

9. Specific objective: Determine resistance of commercial corn hybrids to maize weevils.

Progress FY98: Found large differences in number of progeny produced, development time, and progeny weight of weevils developing on commercial corn hybrids.

Plan of Work for FY99: Complete chemical analyses of corn kernels, and correlate with biological parameters of weevils.

10. Specific objective: Determine resistance of commercial oat cultivars to storage insect pests.

Progress FY98: Found large differences in number of progeny produced and development time of insects developing on commercial oat cultivars. Some cultivars were almost totally resistant to storage insect pests.

Plan of Work for FY99: Correlate chemical and physical properties of oat cultivars with biological parameters of insects.

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RESEARCH PROGRESS AND PLAN

CRIS No. 5430-43000-015-00D

Title: Biological Control of Stored Product Insects With Parasites, Predators and Entomopathogens

Main Objectives:

The goal of this project is to develop economically viable pest management systems that capitalize on desirable biochemical, behavioral, and biological traits of parasites, predators and entomopathogens of stored grain insect pests. Specific objectives include characterizing life histories of promising parasites and predators exposed singly and in combination to single- and multiple-species combinations of stored product pests; identifying the semiochemical and behavioral mechanisms used by these biological control agents to locate, recognize and kill their hosts; characterizing behavioral and semiochemical responses by pests that ameliorate the effectiveness of biological control agents; assessing specific stress factors such as temperature, relative humidity, desiccants, and inhibitors of insect-specific hormones that might weaken pests and make them more susceptible to biocontrol agents; determining parasite-host release ratios, timing of releases, and effect of multiple-species releases on control efficacy; evaluating possible detrimental effects of releasing biological control agents into stored grain commodities; elucidating the biochemical mechanisms by which the toxins of *Bacillus thuringiensis* (*B.t.*) kill stored product pests, with particular emphasis on the physiology of membrane receptor-protein interactions; evaluating the extent of resistance and cross-resistance to native, cloned, and truncated *B.t.* toxins in stored product insect populations; and characterizing insect resistance mechanisms involving the interaction of *B.t.* toxin proteins with midgut proteinases in *Plodia interpunctella*.

Investigators: **Ralph W. Howard, Brenda S. Oppert , Jeffrey C. Lord**

1. Specific Objective: Determine the mechanisms by which *Cephalonomia waterstoni* and *C. tarsalis* recognize their specific hosts, the rusty grain beetle and the saw-toothed grain beetle, respectively.

Progress FY98: Conducted detailed behavioral analyses and generated an ethogram for host recognition by *C. tarsalis*. Manuscript is now published.

Plan of Work for FY99: Continue with development of ethograms for *C. waterstoni*. Continue development of bioassays for chemical cues used in host recognition.

2. **Specific Objective:** Evaluate the possible roles of pheromones in sexual behavior by the parasitoids *Cephalonomia tarsalis* and *Pteromalus cerealellae*.

Progress FY98: Video protocols were established for generating ethograms, behavioral data were gathered, and bioassays were developed.

Plan of work for FY99: Conduct detailed behavioral analyses of the courtship and copulatory behavior of the parasites. Begin characterization of relevant life history parameters that influence pheromone biology.

3. **Specific Objective:** Characterize the cuticular hydrocarbons of the parasitoid *Pteromalus cerealellae* and determine their semiochemical functions.

Progress FY98: Completed identification of all hydrocarbons. Completed statistical analyses of compositional differences.

Plan of Work for FY99: Develop bioassays to characterize semiochemical functions of the hydrocarbons. Begin writing manuscript.

4. **Specific Objective**: Identify cues by which parasitoids locate grain stores containing stored product insects.

Progress FY98: A wind tunnel bioassay chamber was further tested in preliminary experiments using the parasite *C. tarsalis* and its host, the saw-toothed grain beetle.

Plan of Work for FY99: Construct a wind tunnel bioassay chamber specific for parasitoids and begin experiments with four stored product parasitoids, assessing their orientation behavior towards odor volatiles from stored grain and stored grain infested with storage pests.

5. **Specific Objective**: Investigate the involvement of insect gut proteinases in the development of resistance .

Progress FY98: In FY 1997, we described *Bacillus thuringiensis(Bt)* resistant strains of the Indianmeal moth, *Plodia interpunctella*, that lacked a major gut proteinase. During FY98, P. interpunctella larvae collected from different geographic locations were analyzed for gut proteolytic patterns. Results indicated variation in some proteinase patterns, which may help to formulate more effective insect control strategies based on *Bt* toxins. This work was presented at several regional and national meetings, and a manuscript is being prepared for submission to Archives of Insect Biochemistry.

Plan of Work FY99: Evidence from previous studies indicate that beetle gut proteinases process *Bt* toxins similar to moths. Therefore, we will investigate the relationship of *Bt* toxicity and digestive proteinases in beetles. If proteinases are found to be positively correlated to toxicity in beetles, *Bt* toxins may be modified to enhance their efficacy against stored product beetle pests.

6. **Specific Objective**: Determine the nature of interactions among the pathogens *Beauveria bassiana* and *Mattesia dispora*, the parasitoid *Cephalonomia tarsalis,* and their host, the sawtoothed grain beetle.

Progress FY98: This is a new project

Plan of Work FY99: Choice and no-choice tests will be carried out to determine if parasitoids avoid diseased hosts and, if not, whether the diseases are transmitted to and by them.

7. **Specific Objective**: Quantify the consequences of *Bacillus thuringiensis* (*Bt*) resistance development in pest moths, in terms of disease susceptibility.

Progress FY98: This is a new project

Plan of Work FY99: Laboratory bioassays will be conducted against *Bt*-resistant and *Bt*-susceptible strains of the Indianmeal moth and almond moth with *Beauveria bassiana, Metarhizium anisopliae* and *Mattesia dispora*.

8. **Specific Objective**: Determine the role of eicosanoids in the evasion of host immune response by *Beauveria bassiana*.

Progress FY98: This is a new project

Plan of Work FY99: Conduct in vivo tests of immune response to the immuno-suppressive and immuno-inductive propagules of *Beauveria bassiana* in combination with eicosanoid inhibitors or precursors.

9. **Specific Objective**: Determine the extent and nature of synergy between insecticidal silicaceous dusts and *Beauveria bassiana*.

Progress FY98: True synergy was discovered between the entomogenous fungus *Beauveria bassiana* and diatomaceous earth was found in assays against the lesser grain borer.

Plan of Work FY99: Test other target Coleoptera and Lepidoptera and silicaceous materials. Examine cuticle effects of silicaceous materials by light and electron microscopy and by profiles of cuticular hydrocarbons in order to correlate with enhancement of fungus efficacy.

Publications:

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Stanley, D. W., and R. W. Howard. 1998. The biology of prostaglandins and related eicosanoids in invertebrates: Cellular, organismal and ecological actions. Amer. Zool. 38:369-381.

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RESEARCH PROGRESS AND PLANS

CRIS No. 5430-43000-016-00D

Title: Development of Physiological and Genetic Controls for Stored Product Insects

Main Objectives:

The goals of this project are to characterize physiological and genetic processes that can be manipulated for insect control purposes, identify inhibitory proteins and genetically modify cereal grains and entomopathogens with their genes, develop techniques for genetic manipulation of insects, devise genetic methods for managing pesticide resistance in insects and for enhancing the efficacy of beneficial insects. Specific objectives include identifying target sites in insect skeletal, gut and endocrine systems vulnerable to biopesticides; characterizing molting and digestive enzymes, identifying inhibitors whose genes are amenable to plant and microbial genetic engineering; evaluating recombinant seeds for resistance to insects and evaluating recombinant microbial pathogens for efficacy as biological control agents; identifying inhibitors of insect hormones, evaluating modes of action and efficacy as regulators of growth and physiological processes; characterizing cuticular components of pests and parasites, assessing functional responses to environmental stress; characterizing genetic mechanisms regulating reproduction, development, and pesticide resistance of pest and beneficial insects; and developing techniques for genetically altering, manipulating, and monitoring populations of pest and beneficial insects.

Investigators: **Karl J. Kramer, James E. Baker, Richard W. Beeman**

1. **Specific Objective:** Characterize the biochemical mechanism(s) of malathion resistance in the parasitoid *Anisopteromalus calandrae*.

Progress FY98: A purification method based on HPLC and PAGE was developed to purify the malathion-specific carboxylesterase from the resistant strain of *A. calandrae*.

Plan of Work for FY 99: Obtain internal sequence, subsequent to digestion, for amino acid sequence and subsequent design of primers.

2. Specific Objective: Characterize molecular genetics of malathion resistance in *Anisopteromalus calandrae*.

Progress FY98: Carboxylesterase-like enzyme cDNAs have been cloned from resistant and susceptible strains of *A. calandrae*. Sequences have been compared and a single base substitution documented. This substitution leads to an amino acid change in the predicted protein sequence from tryptophan 220 in susceptible strain to glycine 220 in the resistant strain. Northern blot analysis demonstrated that expression levels of the carboxylesterase-like enzyme mRNA and total RNA in adult parasitoids are approximately 30-fold higher in the resistant strain compared with those in the susceptible strain. Southern analysis indicated that Pst I or Eco RI restriction sites are different in the two strains, suggesting a modified gene structure or gene amplification in resistant parasitoids.

Plan of Work for FY99: Develop bacterial expression system for the carboxylesterase.

3. Specific Objective: Initiate biochemical studies on malathion resistance in the Dickinson strain of *Habrobracon hebetor.*

Progress FY98: Detoxification systems were evaluated in susceptible and resistant strains of *H. hebetor*. Esterase patterns as determined by PAGE were significantly different between the two strains.

Plan of Work for FY99: Prepare manuscript describing results.

4. Specific Objective: Initiate studies on proteinases in the lesser grain borer.

Progress FY98: Biochemical and molecular studies were conducted. The effect of a range of inhibitors on chymotrypsin and trypsin activities were determined. Trypsin-like and chymotrypsin-like enzyme cDNAs were cloned and sequenced.

Plan of Work for FY99: Continue biochemical and molecular studies with selected coleopterans.

5. Specific Objective: Determine effect of diatomaceous earth formulations on parasitoids associated with stored grain.

Progress FY98: Three types of bioassays were conducted to evaluate the effect of Protect-It, a formulation containing 90% diatomaceous earth and 10% silica aerogel. This DE formulation had a detrimental effect on efficacy of the parasitoid.

Plan of Work for FY99: Prepare manuscript describing results.

6. Specific Objective: Evaluate NIR as a physical method for species identification of stored product beetles.

Progress FY98: NIR was successfully used as a taxonomic tool for stored product beetles and also could successfully classify parasitized insects in wheat kernels.

Plan of Work for FY99: Evaluate NIR as a classification system for critical biological processes in insects.

7. Specific Objective: Develop a capillary electrophoresis method for the analyisis of phenolic acids in corn.

Progress FY98: A capillary electrophoresis method was developed to quantify phenolic acids in corn. Total phenolics and bound phenolics were determined in corn cultivars and the concentrations of these biochemicals related to resistance levels in the cultivars to the maize weevil.

Plan of Work for FY99: Expand these studies to include more corn cultivars.

8. Specific Objective: Develop methodology for genetic manipulation of pest insects.

Progress FY98: The entire Tribolium white-eye gene and cDNA was cloned using 3' and 5' RACE (Rapid Amplification of cDNA Ends).

Plan of Work for FY99: Work is underway to develop and test white-based gene transfer vectors. Expected benefits include the ability to infect pest species with insect control genes (viruses, lethal genes, transposons), transfer of pesticide resistance genes to parasitoids, and transfer of deleterious traits (such as disease susceptibility, pesticide susceptibility, and cold intolerance) to pest species.

9. Specific Objective: Harness natural insect control genes.

Progress FY98: Interaction of the hybrid incompatibility gene H with the naturally-occurring killer gene M1 was further characterized. Map-based cloning of several genes involved in the M/H interaction was initiated.

Plan of Work for FY99: We will continue to pursue the map-based cloning of other killer genes and incompatibility genes using the technique of Amplified Fragment Length Polymorphism Analysis (AFLP). Understanding the lethal mechanism of these unusual genes could lead to new approaches for pest control.

10. Specific Objective: Identify potential biopesticides and insect growth regulators and screen them for ability to disrupt stored-product insect development and physiology.

Progress FY98: Avidin, an antinutritional protein present in hen's egg white, was found to exhibit excellent biopesticidal activity in transgenic corn.

Plan of Work for FY99: Continue studies on the development of biopesticides and insect growth regulators that disrupt insect gut and cuticle physiology for control of insect pests in food and stored products.

11. **Specific Objective:** Characterize reactants, intermediates, products and enzymes involved in insect cuticle sclerotization and immune responses, which might be targets for novel insect selective control agents.

Progress FY98: We used biochemical and genetic approaches to investigate tyrosine metabolism for cuticle tanning in moth, beetle, fly, and cockroach species, and identified many novel metabolites including phenolic conjugates, catecholamines, and cuticular protein cross-linking agents. These pioneering results added to the scientific knowledge base about insect growth and development, and pinpointed unique metabolic reactions that might be inhibited by novel insect growth regulators or biocontrol agents. Some of the novel metabolites characterized also play critical roles in both the nervous and immune systems of many types of animals. More than fifty years after Pryor in England first proposed the involvement of proteins and phenolic compounds in insect cuticle stabilization, we confirmed this hypothesis and advanced our understanding of the mechanism of cuticle sclerotization, a process that is a potential target for novel insect growth regulators and biopesticides.

Plan of Work for FY99: Continue to identify and characterize insect-specific reactions that occur during cuticle sclerotization and immune responses, which could serve as targets for new insect growth regulators and biopesticides for use in protecting food and stored products against insect pests.

12. **Specific Objective:** Clone insect chitinolytic enzyme genes and study their molecular biology to determine how insects regulate chitin degradation during molting. Use chitinolytic enzymes as biopesticides to help control pest insects by manipulating chitinolytic enzyme genes in transgenic plants and microbial entomopathogens.

Progress FY98: Progress was made in developing a new type of transgene, an insect chitinolytic enzyme gene, as a biopesticide in genetically engineered plants, which should enhance resistance to pest insects. Chitinolytic enzymes degrade chitin, a carbohydrate biopolymer present in the gut lining and exoskeleton of insects. Our results provided the strongest evidence to date that an insect chitinase transgene enhances insect resistance when expressed in plants. Tobacco plants expressing insect chitinase were more resistant to the tobacco budworm than were plants that did not express the enzyme. This study also contributed knowledge at the organismal and molecular levels about chitinolytic enzyme-insect-plant interactions and was crucial for the development of transgenes such as insect chitinolytic enzyme genes for insect control purposes in host plant resistance genetic engineering programs. A patent was issued for the use of this enzyme as a biocide for insect control applications.

Plan of Work for FY99: Continue to develop insect chitinolytic enzymes as biopesticides for use in protecting food and stored products from insect pests and microbial pathogens.

Publications:

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Patent:

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RESEARCH PROGRESS AND PLANS

CRIS No. 5430-43000-017-00D

Title: Monitoring and control strategies for stored-product insects

Main objectives:

The overall objective of this CRIS is to develop monitoring and control strategies for stored-product insect pests in and around storage and processing facilities. These studies will lead to the development and improvement of genetic, semiochemical, and physical techniques to monitor insect behavior, estimate population densities, and determine control thresholds and timing of management actions. Selected control tactics will be evaluated for integration into ecologically-compatible pest management programs. Insect resistant packaging and barriers will be developed to prevent infestation of stored products. The acquired knowledge will lay the ground work necessary to evaluate the efficacy of current practices and to develop ecologically-sound insect pest management programs.

Investigators: **Michael A. Mullen, Franklin H. Arthur, Alan K. Dowdy**

1. **Specific Objective:** Development of a disposable pheromone baited trap for stored-product Coleoptera.

Progress FY 98: The trap was developed from a 15 dram snap cap vial with openings for the insects to enter and baited with a pheromone lure. A food oil was used as a short range attractant and a killing agent. Laboratory comparisons with the FLIT TRAK M^2 that showed the vial trap to be less effective for capturing adult red flour beetles. This trap was designed as a special use trap to be used in areas where the other traps would not be suitable.

Plan of Work for FY 99: Research has been completed and a manuscript has been accepted.

2. **Specific Objective**: Develop an oil base food attractant for stored product beetles.

Progress FY 98: Extractions were made from corn and tested for attractiveness by comparing them to the current wheat-oat-sesame oil blend. The corn oil blend was generally found to be more attractive, however, there are indications that extractions made from different corn varieties varied in their attractiveness.

Plan of Work for FY 99: Tests will be continued using different corn varieties as well as other oil seeds. Different extraction methods will be compared.

3. Specific Objective: Redesign the FLIT-TRAK M^2 to improve the rate of catch and ease of counting trapped insects.

Progress FY 98: It became apparent that a wall mounted version of the FLITe-TRAK was not feasible. A new wall mounted trap was developed to complement the floor mounted pitfall design. Several trap designs have been developed and are currently being tested.

Plan of Work for FY99: As new prototypes become available, testing will be continued.

4. Specific Objective: A new pheromone trap will be designed for use in specialized areas, such as grocery stores, so that they will remain out of the view of the public.

Progress FY 98: An under the shelf trap was designed and tested in grocery stores. The trap was found to be as effective as the Pherocon II.

Plan of Work FY99: A manuscript has been prepared and several companies are interested in marketing of the trap.

5. Specific Objective: Use of pheromone traps as a dispersal mechanism for granulosis virus.

Progress FY 98: No transmission of the virus using pheromone baited traps was demonstrated and the project has been terminated.

6. Specific Objective: Improvement of seals and closures on commercial packaging to reduce infestation by stored-product insects.

Progress FY 98: Cooperation with food processing companies was expanded. Tests for several companies were conducted. A new package was developed and is now undergoing testing.

Plan of Work for FY 99: Testing of packages will continue and recommendations will be made to the manufacturers to improve insect resistance.

7. Specific Objective: Test chemical odor neutralizers for use in insect resistant packaging.

Progress FY98: Testing was terminated.

Plan of Work for FY 99: Test on odor neutralizers has been discontinued and no further work is planned.

8. Specific Objective: Test, evaluate, and make recommendations on several new repellant compounds for use in insect resistant packaging.

Progress FY 98: These compounds were tested on breakfast cereal, cat food , and baby cereal.

One repellant, Repelkote (methyl salicylate) was approved for use on packages by the FDA. Other compounds were screened for effectiveness as repellants, and two were identified as effective in laboratory tests. Longevity tests indicated that one compound was effective for six months. Full scale package tests are now being conducted.

Plan of Work FY99: As new compounds become available they will be tested. Full scale package tests will be continued.

9. Specific Objective: Determine sources of insect infestation along marketing channels by using DNA fingerprinting technology.

Progress FY98: Continued analysis of lesser grain borer to identify genetic markers that can differentiate individual insects into population groups. More than 300 primers have been tested on 13 lesser grain borer populations. Data analysis is in progress.

Plan of work FY99: Work will continue to identify genetic markers. New molecular biological techniques will be employed. Initiate a project to study seasonal changes in the genetic profile of insect populations in farm-stored grain.

10. Specific Objective: Examine DNA fingerprints to detect and monitor insecticide resistance.

Progress FY98: Screening of Indianmeal moth for genetic markers that are linked to resistance to the toxin produced by *Bacillus thuringiensis*. Initiated a project to study resistance to phosphine in lesser grain borers.

Plan of work for FY99: Research on using DNA fingerprints to detect and monitor insecticide resistance in the Indianmeal moth, lesser grain borer, and flat/rusty grain beetle will continue. This may lead to a quick test to be used in field situations.

11. Specific Objective: Evaluate the distribution of heat in cereal grain processing plants as an alternative to methyl bromide fumigation for insect control.

Progress FY98: Research was conducted in three commercial processing facilities. Generally adequate insect control occurs throughout the heated areas. Within a room, however, there are cool pockets where insects survived and hot pockets that were overheated and may cause damage to processing equipment. Air circulation during the heat treatment is important to facilitate even heating and equilibrate the cooler and warmer areas. This will result in better utilization of heating energy and improved insect management.

Plan of work for FY99: Research will be conducted to evaluate the penetration of heat into processing equipment. We will begin development of an automated temperature monitoring system to improve the application of heat and minimize worker safety issues.

12. Specific Objective: Evaluate the combined use of heat and diatomaceous earth on insect mortality.

Progress FY98: In the laboratory, four diatomaceous earth formulations were evaluated to determine which was most efficacious in combination with heat treatments. Exposure to the Protect-It® formulation caused greater beetle mortality than any of the other materials. Withholding food from the beetles after exposure to heat plus diatomaceous earth increased mortality 2 to 4 times beyond the level for beetles that were not denied food. A combination treatment of heat with an effective diatomaceous earth formulation may be useful for managing this pest in cereal-grain processing facilities. A full mill treatment of heat plus diatomaceous earth was applied in the pilot flour mill at Kansas State University. In areas of the mill where temperatures were in excess of 47°C for at least 25 h, diatomaceous earth applications in combination with the heat were no more effective than the heat treatment alone. Where less heating occurred, beetles exposed to diatomaceous earth died sooner and faster than insects not exposed to the insecticidal dust. These results indicate that the application of diatomaceous earth in areas that cannot be heated to 47°C is effective for controlling *T. confusum* in a flour mill.

Plan of work for FY99: Research is planned to better understand the time, temperature, and humidity requirements necessary for mortality of several stored-product insects.

13. Specific Objective: Evaluation of residual insecticides on different flooring surfaces.

Progress FY98: Red flour beetles were exposed for 15-120 minutes on concrete treated with cyfluthrin wettable powder, removed, and placed on filter paper with flour or with no flour, and held for one week to assess survival. Bioassays were conducted every 2 weeks for 16 weeks. Knockdown increased as the exposure interval increased, and decreased at each interval as residues aged. Until week 6, < 5% of the beetles survived when exposed for > 30 minutes. After week 6, survival began to increase at exposure intervals > 30 minutes and by week 16, survival usually exceeded 50%. In contrast, beetles recovered from knockdown and survived when given flour. Survival on flour exceeded 80% by week 6 at all exposure intervals.

Plan of work for FY99: Cooperate with industry to conduct evaluations of new chemicals. Conduct additional research with established chemicals.

14. Specific Objective: Determine residual efficacy of new insecticides as grain protectants.

Progress FY98: Hard red winter wheat was treated with encapsulated cyfluthrin, stored at 20, 25, 30, and 35 °C. for 8 months, and bioassayed at bi-monthly intervals with adult rice weevils. Weevil survival and the number of F_1 adults was not related to temperature but was inversely dependent on concentration, and increased as residues aged. An application rate of at least 2 ppm would be necessary to control rice weevils for 10 months on stored hard red winter wheat.

Plan of work for FY99: Evaluate new formulations of diatomaceous earth for use on stored grains.

15. Specific Objective: Determine the potential for the expanded use of aeration on stored grains.

Progress FY98: Population growth models were integrated with temperature data to predict insect development at different aeration management strategies for hard red winter wheat stored in the southern plains.

Plan of work for FY99: Continue cooperative efforts regarding insect population models and how they could be used to develop aeration plans.

16. Specific Objective: Evaluate new biorational chemicals as protectants of stored grains.

Progress FY98: Tests with several natural products have been completed. None of these chemicals controlled stored-product insects.

Plan of work for FY99: Continue with research regarding promising candidate microbial and fungal pathogens.

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